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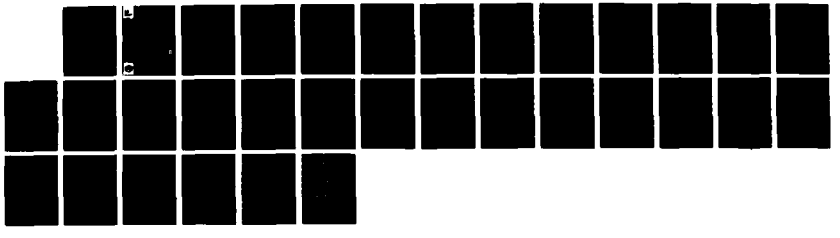

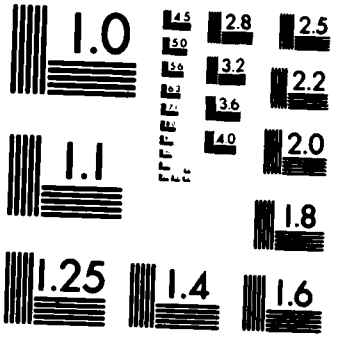

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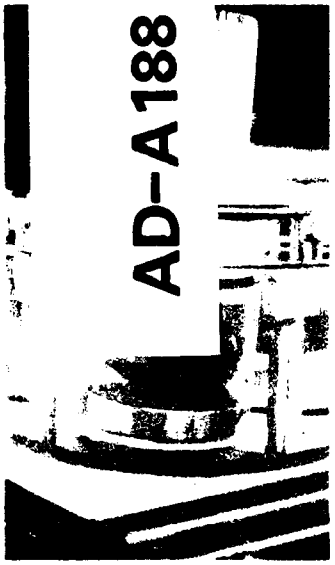

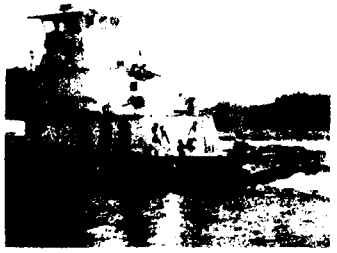

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ENVIRONMENTAL EFFECTS OF NAVIGATION TRAFFIC: LABORATORY STUDIES OF THE EFFECTS ON MUSSELS OF INTERMITTENT EXPOSURE TO TURBULENCE AND SUSPENDED SOLIDS

by

Barry S. Payne, Andrew C. Miller

Environmental Laboratory

DEPARTMENT OF THE ARMY
Waterways Experiment Station, Corps of Engineers
PO Box 631, Vicksburg, Mississippi 39180-0631

and

David W. Aldridge

Department of Biology
North Carolina A&T State University
Greensboro, North Carolina 27411

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Navigation traffic can intermittently increase turbulence and suspended solids in rivers. Freshwater mussels are essentially sessile filter-feeders and could be susceptible to physiological disruption as a result of exposure to these potential effects of navigation traffic. In laboratory studies, cyclic increases in water velocity and turbulence at a level and frequency that can be caused by routine navigation traffic led to reduced feeding rates and slightly increased reliance on endogenous and nonproteinaceous energy reserves. However, continuous or near-continuous disruption, such as might be associated with barge fleeting activities, led to virtually complete reliance on endogenous energy reserves. These results suggest that proposed areas of barge fleeting be evaluated on a site-specific basis to ensure that important mussel populations are not situated in nearby habitats that will be exposed to sustained periods of turbulence and suspended solids above ambient conditions. In addition, these laboratory studies confirm the utility of sublethal physiological indices of stress, such as oxygen:nitrogen and flesh:shell mass ratios, in field studies aimed at monitoring environmental impacts on natural mussel populations.

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PREFACE

In October 1982 the US Army Engineer Waterways Experiment Station (WES) initiated a study of the effects of navigation traffic in large waterways, as part of the Environmental and Water Quality Operational Studies (EWQOS) Program. The purpose of this research was to analyze physical and biological effects of commercial navigation traffic using results of field and laboratory experiments and a synthesis of the available literature. This research has been continued since October 1985 under the Environmental Impact Research Program (EIRP), Work Unit 32393. Additional support for these studies has been provided by the Navigation Planning Support Center (NPSC) of the US Army Engineer District, Louisville.

This report, prepared by Drs. Barry S. Payne and Andrew C. Miller, WES, and Dr. David W. Aldridge, North Carolina A&T State University, provides the results of two laboratory experimental studies of physical effects of commercial navigation traffic and discusses these results in relation to field observations. The report was edited by Ms. Jessica S. Ruff of the WES Information Products Division.

The study was conducted under the general supervision of Mr. Richard Coleman, Acting Chief, Aquatic Habitat Group (AHG); Dr. Thomas D. Wright, former Chief, AHG; Dr. C. J. Kirby, Chief, Environmental Resources Division; and Dr. John Harrison, Chief, Environmental Laboratory, WES. Dr. Roger Saucier is Program Manager of the EIRP. Dr. John Bushman and Mr. Earl Eiker of OCE and Mr. Dave Mathis of the Water Resources Support Center were the Technical Monitors. Mr. Terry Seimsen served as Technical Monitor for the NPSC. Dr. Jerome Mahloch was Program Manager of the EWQOS.

Commander and Director of WES was COL Dwayne G. Lee, CE. Technical Director was Dr. Robert W. Whalin.

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CONTENTS

ENVIRONMENTAL EFFECTS OF NAVIGATION TRAFFIC: LABORATORY STUDIES OF THE EFFECTS ON MUSSELS OF INTERMITTENT EXPOSURE TO TURBULENCE AND SUSPENDED SOLIDS

PART I: INTRODUCTION

Background

1. Dense and diverse beds of freshwater mussels often occur in gravelly shoals of large rivers (Miller et al., in preparation). Suitable habitat for such beds in major rivers such as the Ohio, Mississippi, and Tennessee has been seriously reduced by major modifications to control flooding and facilitate commercial navigation. Remaining mussel beds, some of which are harvested for commercial purposes (Miller et al., in preparation) and contain individuals of some of the 28 species protected under the Endangered Species Act (see Federal Register, 50 CFR 17.11 and 17.12, 10 April 1987), deserve and receive special protection as valued natural resources that are now limited in distribution.

2. Filter-feeders such as bivalves are especially sensitive to increased levels of turbulence and turbidity (Widdows, Fieth, and Worral 1979). Considerable concern exists that increased turbulence and turbidity sometimes associated with navigation traffic (Simons et al. 1981) will have detrimental effects on remaining populations of mussels. Most impact work on turbulence and turbidity effects has been carried out on marine bivalves and has involved the continuous exposure of test animals to constant, and often unnaturally high, turbidity levels (Moore 1977, Wilber 1983). Navigation traffic may intermittently expose some freshwater mussels to turbulence and turbidity above ambient levels. Factors such as the proximity to the navigation channel and seasonal exposure to ambient turbulence and turbidity (Simons et al. 1981) determine if or to what degree a specific habitat area is impacted.

3. The major effect of increased levels of turbulence and turbidity on bivalves is to reduce the rate and/or efficiency of feeding (Moore 1977). Unfortunately, evaluating these parameters in the field to assess navigation impacts is very difficult. However, reduced feeding efficiency can result in

long-term physiological changes that can be more easily evaluated in the field. Typically, starving or semistarved invertebrates show changes in metabolic rates (Barnes, Barnes, and Finlayson 1963; Bayne 1973; Logan and Epifanio 1978; Capuzzo and Lancaster 1979; Dawirs 1983; Page 1983) and shifts to alternate catabolic substrates (Ansell and Sivadas 1973; Bayne 1973; Ikeda 1977; Russell-Hunter et al. 1983). Such shifts have been shown to be useful indicators of sublethal environmental stress in molluscs (Widdows 1978; Bayne et al. 1979; Bayne, Clarke, and Moore 1981).

Purpose

4. The purpose of this report is to describe the physiological changes induced in laboratory studies in which freshwater mussels were cyclically exposed to turbulence and turbidity. Two experimental studies are reported. The first measured metabolic rate and catabolic substrate shifts of three species of mussels that were cyclically exposed to unnaturally high levels of turbulence and turbidity at two distinct frequencies. This experiment was designed both to evaluate the importance of frequency of cyclic exposure to physiologically disruptive changes in hydrologic conditions and to assess the utility of food clearance, respiration, and nitrogen excretion rate measurements as quantitative indices of stress.

5. The second experiment dealt with metabolic rate and tissue condition shifts of a single species exposed to continuously low, cyclically high, and continuously high turbulence without increased turbidity. The levels of turbulence exposure used in the second experiment were within the range of measurements that have been made in the field in relation to navigation traffic. Each experiment is individually discussed in relation to relevant published information on physiological adjustments of molluscs to sublethal stress. Then, the implications of these laboratory experimental studies for predictions and field evaluations of the impacts of navigation traffic on natural populations of freshwater mussels are discussed.

PART II: THE EFFECTS OF INTERMITTENT EXPOSURE TO TURBULENCE AND SUSPENDED SOLIDS ON THREE SPECIES OF FRESHWATER MUSSELS

Materials and Methods

6. The three species of mussels used in the experiment, *Quadrula pustulosa* (Lea), *Fusconaia cerina* (Conrad), and *Pleurobema beadleanum* (Lea) (Order Schizodonta: Family Unionidae), represent genera that occur throughout the Mississippi River drainage and many other rivers that drain into the Gulf of Mexico. Clams were collected on 5 July 1983 from two adjacent riffles in the Tangipahoa River in southwestern Mississippi. On 5 July, the water at the site had a temperature of 21° C, calcium hardness of 3.2 mg/ℓ divalent calcium, pH of 6.8, and turbidity of 8.9 NTU. In general, the Tangipahoa is a clear, rapid-flowing river (Miller, Payne, and Aldridge 1986). All clams were collected from gravelly sands at water depths ranging from 0.25 to 1.0 m. Approximately 200 individuals were collected and brought immediately to the laboratory.

7. On 6 July, clam shells were cleaned of aufwuchs and gradually introduced to aerated and dechlorinated Vicksburg city tap water (well source: 18 mg/ℓ divalent calcium) and maintained at 20° C. During both this laboratory acclimation period and subsequent experiments, clams were fed a suspension of brewer's yeast. On 9 July, an identification number was scratched on the shell of each clam using a carbide scribe.

8. The 52 *Q. pustulosa*, 68 *F. cerina*, and 50 *P. beadleanum* were then divided into four groups per species of approximately equal number and size distribution. Beginning on 9 July (day 0), these groups were subjected to one of four treatments:

a. Treatment 1, Infrequent turbulence and suspended solids. Clams were exposed to suspended solids (average maximum value of 750 mg/ℓ) created by low levels of turbulence maintained for 7 min every 3 hr.

b. Treatment 2, Infrequent turbulence. This was a control for the previous experimental treatment in which the clams were exposed to low levels of turbulence but not suspended solids for 7 min every 3 hr.

c. Treatment 3, Frequent turbulence and suspended solids. Clams were exposed to suspended solids (average maximum value of 600 mg/ℓ) created by low levels of turbulence maintained for 7 min every 0.5 hr.

d. Treatment 4, Frequent turbulence. This was a control for the third experimental treatment in which the clams were exposed cyclically to low levels of turbulence but not suspended solids for 7 min every 0.5 hr.

9. The four groups were exposed to their respective treatments in glass aquaria (25 × 51 × 20 cm) containing 30 ℓ of constantly but gently aerated water. Two tanks were used for each group to prevent crowding and to replicate each treatment. The turbulence necessary to cyclically suspend the solid material (diatomaceous earth) in each experimental tank was produced by two centrifugal water pumps (Aqualogy model 10-120). The pumps stayed on for 5 min and were 2 min out of phase with each other to keep diatomaceous earth from permanently settling on quiescent areas on the bottom of the tanks. On-off times for the pumps were controlled by an electronic timer.

10. The diatomaceous earth was washed seven times before being added to the tanks to remove most of the very fine particles (not settled in 30 min). Thus, solids to be resuspended consisted largely of intact diatom tests. Peak concentrations of suspended solids in samples from all tanks were monitored using a Bausch and Lomb model 710 spectrophotometer at a fixed wavelength (550 μ) so that diatomaceous earth cleared by clams could be determined by comparing sample absorption to an appropriate standard curve.

11. Following turbulence, the suspended solid concentrations in treatments 1 and 3 reached their peak of 750 and 600 mg/ℓ, respectively, and fell to 10 percent of that peak value within 15 min after the end of turbulence. Mininum suspended solids concentrations for treatments 1 and 3 were 25 and 125 mg/ℓ, respectively. The inability to get zero levels of suspended solids between successive episodes of turbulence is a general problem in turbidity studies (Moore 1977). The peak suspended solids concentrations used in this study would be expected only in areas affected severely by dredging and navigation impacts (Wilber 1983; Robinson, Wehling, and Morse 1984).

12. To evaluate the food ingestion rates of the 170 clams, their ability to clear yeast from a suspension of known concentration was evaluated at 20° C on day 4. Each clam was placed in 100 ml of water to which the equivalent of 8 mg of dry yeast was added volumetrically from a premixed suspension. Clams were allowed to filter for 1 hr. More yeast concentration was added if, during this hour, the concentration clearly dropped below half the starting value. After 1 hr, each clam was removed and returned to its respective tank. The final yeast concentrations were determined by measuring

optical densities at 550 μ using the spectrophotometer. Reductions in concentrations relative to controls (yeast suspensions without clams) were easily converted to a food clearance rate (milligrams per gram per hour) using an empirical relationship of the optical density to yeast concentration. Obviously, feeding clams yeast suspensions in a closed chamber is not fully representative of natural conditions. Nevertheless, our measurements of food clearance rates provide relative estimates of the feeding state of the clams in the different treatments.

13. To evaluate levels of metabolic activity of the clams, individual oxygen uptake rates were monitored for day 5 through day 8 with Clark-type polarographic oxygen electrodes using a Yellow Springs Instrument model 5302 respirometer. Water in the respirometer was stirred by a magnet gently spinning below a mesh screen supported by a stainless steel annulus. Clams were placed on the screen above the magnet. Approximately 50 ml of water without yeast or suspended inorganic solids was used in the respirometer. In all cases, oxygen uptake rates were measured on clams with their siphons opened normally. Uptake rates were monitored from 90- to 65-percent air saturation in water at 20° C. Clams spent 20 to 30 min in the respirometer chamber. A few clams were too large to fit in the respirometer. Thus, oxygen uptake rates were not measured for 9 *Q. pustulosa* and 10 *F. cerina* from all treatments.

14. To detect major shifts in catabolic substrates (proteins versus carbohydrates and fats), the total nitrogen (ammonia plus urea) excretion rate for each clam was assessed on day 9. Clams were placed individually in 50 or 100 ml of water, depending on their size. At the end of 1 hr the clams were removed, and 3 mg Fisher U-21 urease was added for every 50 ml of water. Total ammonia excreted as ammonia or urea was then determined using an Orion model 95-10 ammonia probe coupled to an Orion Model 407 A/L specific ion meter, following methods described in Russell-Hunter et al. (1983). All of the nitrogen excretion rates were measured during a 1-hr period on day 9 because nitrogen excretion is typically more labile than oxygen uptake (Bayne 1973; Aldridge 1983; Russell-Hunter et al. 1983; Aldridge, Russell-Hunter, and Buckley 1986); hence, this procedure results in reliable oxygen: nitrogen (O:N) ratios for comparing treatment effects.

15. Individual clams were never kept out of their treatment tanks for more than 90 min during any one of the three physiological rate

determinations. On day 10, all clams were sacrificed by shucking the flesh from the shells. Prior to weighing the soft tissues were dried to constant weight for 2 days at 75° C.

Results and Discussion

16. Three paired comparisons of treatments were made. First, the effect of the periodicity of two levels of physical disturbance (turbulence) was assessed by comparing the physiological condition of clams in the infrequent versus frequent turbulence treatments (2 and 4). In the other two comparisons, the combined effects of suspended solids and turbulence exposure were evaluated by comparing treatments 1 and 2 as well as 3 and 4. No differences between replicate aquaria were seen ($p > 0.05$, two-tailed t-test).

17. While acknowledging that there is no zero-level control for assessing turbulence effects, a comparison of treatments 2 and 4 is useful in assessing the relative effects on clams of varying the time interval between brief periods of turbulence (Table 1). All three species responded to more frequent turbulence by lowering nitrogen excretion rates and, hence, increasing O:N. Values for O:N were derived from each pair of rate determinations made on an individual clam.

18. The O:N ratio provides an assessment of the relative contribution of protein to total catabolism (Corner, Cowey, and Marshall 1975; Ikeda 1977; Widdows 1978; Bayne and Newell 1983; Russell-Hunter et al. 1983). Protein-based catabolism is indicated by O:N values less than 30 (Bayne and Widdows 1978). Infrequent exposure to turbulence had no major effect on the clams. All three species yielded O:N values averaging 13, which reflected their ability to base their metabolism on almost total reliance on the proteinaceous food ration of yeast. Only *P. beadleanum* showed higher values of O:N with more frequent exposure to turbulence. This species showed a change in O:N from 11 to 62, reflecting a shift to catabolism based mainly on nonproteinaceous body stores.

19. The effects of infrequent exposure to elevated suspended solids were evaluated by comparing them to the effects of infrequent exposure to turbulence alone. When exposed infrequently to suspended solids and turbulence (treatment 1), all three species showed significant and substantial reductions in food clearance rates. In addition, *G. pustulosa* and *P. beadleanum* showed

Table 1

Tissue Dry Weights, Food Clearance Rates, Oxygen Uptake Rates, Nitrogen Excretion Rates, and O:N Ratios ($\bar{X}$ ± SD) for Mussels Exposed to Infrequent and Frequent Turbulence

Species	Physiology Monitor*	Treatment: Infrequent Turbulence	Treatment: Frequent Turbulence	Student's t-Test: df	Student's t-Test: t	Student's t-Test: Significance
Q. pustulosa	TDW	1.05 ± 0.23	1.13 ± 0.44	22	1.24	NS**
	FCR	7.86 ± 5.63	8.93 ± 7.00	24	0.43	NS
	$\dot{V}O_2$	18.20 ± 5.75	15.52 ± 4.16	20	1.25	NS
	NE	2.61 ± 0.78	1.66 ± 0.61	24	3.46	$p < 0.01$
	O:N	14.00 ± 4.38	17.22 ± 4.22	20	1.76	NS
F. cerina	TDW	0.98 ± 0.37	1.02 ± 0.36	32	0.33	NS
	FCR	11.56 ± 8.80	8.00 ± 3.59	32	1.54	NS
	$\dot{V}O_2$	16.82 ± 5.62	14.81 ± 3.86	27	1.13	NS
	NE	2.42 ± 1.18	1.49 ± 0.80	32	2.63	$p < 0.05$
	O:N	13.86 ± 4.04	32.42 ± 38.67	27	1.79	NS
P. beadleanum	TDW	0.36 ± 0.07	0.36 ± 0.09	28	0.16	NS
	FCR	7.92 ± 8.83	9.71 ± 7.99	28	0.59	NS
	$\dot{V}O_2$	20.81 ± 6.41	22.02 ± 9.80	28	0.13	NS
	NE	3.96 ± 1.11	2.17 ± 1.85	28	3.16	$p < 0.01$
	O:N	11.30 ± 4.73	62.34 ± 92.78	28	2.05	$p < 0.05$

* TDW = tissue dry weight (g); FCR = food clearance rate (mg yeast $\cdot g^{-1} \cdot hr^{-1}$); $\dot{V}O_2$ = oxygen uptake rate (µmol $O_2 \cdot g^{-1} \cdot hr^{-1}$); NE = nitrogen excretion rate (µmol N $\cdot g^{-1} \cdot hr^{-1}$).

** NS = not significant at $p > 0.05$ level.

reduced oxygen uptake and nitrogen excretion rates (Table 2). However, shifts in O:N were not observed.

20. The combined effects of suspended solids and turbulence exposure were more severe at high frequencies of exposure (treatment 3). All three species showed significant reductions in food clearance and nitrogen excretion rates (Table 3). Major shifts in O:N were made by all of the clams exposed to frequent turbidity, to the extent that their catabolism had become entirely based on nonproteinaceous body stores as indicated by O:N ratios in excess of 145.

21. Exposure of all three species of unionid mussels to infrequent (once every 3 hr) and frequent turbidity (once every 0.5 hr) at levels of 750 and 600 mg/ℓ, respectively, caused reduced food clearance rates. Frequent exposure to turbidity resulted in reduced nitrogenous excretion rates in all three species and higher O:N ratios. The response to infrequent exposure to turbidity was more variable, with only *Q. pustulosa* and *P. beadleanum* showing major responses. Both oxygen uptake and nitrogenous excretion decreased rates in tandem. The fact that the animals exposed to turbidity infrequently showed no shift in catabolic substances (O:N ratio) suggests that they were less seriously affected than mussels exposed frequently to turbidity.

22. These findings of reduced food clearance rates of food particles by freshwater mussels exposed intermittently to high concentrations of suspended solids are supported by work on the bivalves *Crassostrea virginica* (Loosanoff and Tommers 1948), *Mytilus edulis* (Widdows, Fieth, and Worral 1979), and *Spisula solidissima* (Robinson, Wehling, and Morse 1984) as well as the filter-feeding gastropod *Crepidula fornicata* (Johnson 1971). Work by both Widdows, Fieth, and Worral (1979) and Robinson, Wehling, and Morse (1984) indicated that concentrations of inorganic suspended solids equaling 100 mg/ℓ can have a major impact on food clearance rates in *M. edulis* and *S. solidissima*. That such reductions in food clearance rates are ultimately translated into reductions in growth rates is seen in the suspended solids research on *Mercenaria mercenaria* (Pratt and Campbell 1956; Bricelj, Malouf, and de Quillfeldt 1984), though not over the very brief 10-day period of the present study.

23. Less work has been done with filter-feeders on the effects that suspended solids have on other aspects of their physiology (e.g., oxygen uptake and nitrogen excretion). However, it appears that imposed starvation or semistarvation is the major impact of high levels of suspended solids and,

Table 2

Tissue Dry Weights, Filter Clearance Rates, Oxygen Uptake Rates, Nitrogen Excretion Rates, and O:N Ratios ($\bar{X}$ ± SD) for Mussels Exposed to Infrequent Turbulence and Infrequent Turbulence Plus Turbidity

Species	Physiology Monitor*	Treatment: Infrequent Turbulence	Treatment: Infrequent Turbulence Plus Turbidity	Student's t-Test: df	t	Significance
M. pustulosa	TDW	1.05 ± 0.23	1.19 ± 0.44	24	1.00	NS**
	FCR	7.86 ± 5.63	3.35 ± 2.74	24	2.76	$p < 0.05$
	$\dot{V}O_2$	18.20 ± 5.75	12.48 ± 4.75	20	2.10	$p < 0.05$
	NE	2.61 ± 0.78	1.51 ± 0.38	24	4.54	$p < 0.001$
	O:N	14.00 ± 4.38	16.34 ± 6.42	20	1.00	NS
F. cerina	TDW	0.98 ± 0.37	0.99 ± 0.29	32	0.06	NS
	FCR	11.56 ± 8.80	5.07 ± 3.51	32	2.82	$p < 0.02$
	$\dot{V}O_2$	16.82 ± 5.62	13.55 ± 4.69	27	1.71	NS
	NE	2.42 ± 1.18	1.73 ± 0.75	32	1.99	NS
	O:N	13.86 ± 4.05	19.40 ± 11.86	27	1.66	NS
M. beadleanum	TDW	0.36 ± 0.07	0.36 ± 0.06	26	0.12	NS
	FCR	7.92 ± 8.83	3.28 ± 1.49	26	2.28	$p < 0.05$
	$\dot{V}O_2$	20.81 ± 6.42	12.15 ± 6.43	26	3.44	$p < 0.01$
	NE	3.96 ± 1.11	2.37 ± 1.06	26	3.90	$p < 0.001$
	O:N	11.30 ± 4.73	15.21 ± 15.87	26	0.88	NS

* TDW = tissue dry weight (g); FCR = food clearance rate (mg yeast $\cdot g^{-1} \cdot hr^{-1}$); $\dot{V}O_2$ = oxygen uptake rate (μmol $O_2 \cdot g^{-1} \cdot hr^{-1}$); NE = nitrogen excretion rate (μmol $N \cdot g^{-1} \cdot hr^{-1}$).

** NS = not significant at $p > 0.05$ level.

Table 3

Tissue Dry Weights, Filter Clearance Rates, Oxygen Uptake Rates, Nitrogen Excretion Rates, and O:N Ratios for Mussels Exposed to Frequent Turbulence and Frequent Turbulence Plus Turbidity

Species	Physiological Monitor*	Treatment: Frequent Turbulence	Treatment: Frequent Turbulence Plus Turbidity	Student's t-Test: df	Student's t-Test: t	Student's t-Test: Significance
Q. pustulosa	TDW	1.13 ± 0.44	1.10 ± 0.27	24	0.17	NS**
	FCR	8.93 ± 7.00	2.36 ± 2.19	24	3.23	$p < 0.01$
	$\dot{V}O_2$	15.52 ± 4.16	13.42 ± 3.75	19	1.22	NS
	NE	1.66 ± 0.61	0.11 ± 0.07	24	9.09	$p < 0.00$
	O:N	17.22 ± 4.22	233.50 ± 69.04	18	9.95	$p < 0.00$
F. cerina	TDW	1.02 ± 0.36	0.97 ± 0.30	32	0.41	NS
	FCR	8.03 ± 3.59	5.12 ± 4.37	32	2.10	$p < 0.00$
	$\dot{V}O_2$	14.82 ± 3.86	15.80 ± 3.73	24	0.71	NS
	NE	1.49 ± 0.80	0.24 ± 0.31	32	6.01	$p < 0.00$
	O:N	32.42 ± 38.67	216.78 ± 112.91	24	6.05	$p < 0.00$
P. beadleanum	TDW	0.36 ± 0.09	0.33 ± 0.07	29	0.76	NS
	FCR	9.71 ± 7.99	3.34 ± 2.74	29	2.76	$p < 0.0$
	$\dot{V}O_2$	22.02 ± 9.80	16.64 ± 4.78	29	1.92	NS
	NE	2.17 ± 1.85	0.23 ± 0.11	29	4.05	$p < 0.0$
	O:N	62.34 ± 92.78	149.19 ± 50.50	28	6.88	$p < 0.0$

* TDW = tissue dry weight (g); FCR = food clearance rate (mg yeast $\cdot g^{-1} \cdot hr^{-1}$); $\dot{V}O_2$ - oxygen uptake rate (μmol $O_2 \cdot g^{-1} \cdot hr^{-1}$); NE = nitrogen excretion rate (μmol N $\cdot g^{-1} \cdot hr^{-1}$).

** NS = not significant at $p > 0.05$ level.

indeed, other environmental stresses on filter-feeders. Generally, the long-term response of most poikilotherms to reduced food availability is to lower metabolic rates (Bayne 1973, Bayne et al. 1979, Russell-Hunter et al. 1983) and to shift to alternative catabolic substrates (Russell-Hunter and Eversole 1976, Widdows 1978, Bayne et al. 1979). Lower oxygen uptake rates are universally an indicator of lower metabolic rates in aerobic organisms (Prosser 1973).

24. Much more variable are the types of shifts seen in the use of alternative catabolic substrates. In some organisms, such as overwintering *M. edulis*, starvation stress shifts the animal from its normal catabolic energy sources of carbohydrates and lipids (high O:N ratios) to a more proteinaceous catabolism (low O:N ratios) (Widdows 1978). In our studies on freshwater mussels, however, the clams exposed to frequent turbidity and turbulence shifted from the control catabolism heavily based on protein (O:N <20) to a catabolism presumably based on stored carbohydrates and lipids (O:N >100), which would ordinarily be used in reproduction or overwintering. Summer O:N ratios for unionids in nature are normally less than 50 (unpublished observations), as are summer O:N ratios for other freshwater molluscs (Aldridge 1985). Such a diversity of responses emphasizes the need for using appropriate controls in assessing the effects of environmental perturbations on the physiological energetics of different types of organisms (Bayne, Clarke, and Moore 1981).

25. In summary, the intermittent exposure of freshwater mussels to high levels of suspended solids (600 to 750 mg/ℓ) disrupted feeding and caused shifts to catabolism of endogenous nonproteinaceous energy reserves. These shifts were obvious from measurements of O:N ratios. Such measurements of responses to intermittent turbidity should be useful in evaluating and managing the ecological impacts of navigation and dredging activities on freshwater mussels.

PART III: THE EFFECTS OF CONTINUOUS AND INTERMITTENT EXPOSURE TO TURBULENCE ON THE FRESHWATER MUSSEL *FUSCONAIA EBENA*

Materials and Methods

26. Seventy-two juvenile *F. ebena*, ranging in shell length from 17 to 26 mm, were collected on the Ohio River (river mile 967), near Olmsted, Ill., on 27 August 1985. The mussels were in a distinct mussel bed that supported a dense and diverse molluscan community (Miller, Payne, and Siemsen 1986). Water depth where mussels were collected ranged from 3 to 5 m. River stage was near the average annual minimum on 27 August. The mussels were brought to the laboratory in Vicksburg, Miss., and gradually acclimated to aged, dechlorinated tap water.

27. On 9 September, the 72 mussels were divided into three groups of approximately equal size distribution. Each group was exposed to one of three conditions: continuous-low, continuous-high, and cyclic-high water velocity. The experiment was conducted in three identical 200-ℓ Plexiglas chambers connected by a central mixing reservoir. The three conditions were created by manipulating the magnitude and duration of velocities of water flowing over gravel in which mussels were positioned (Table 4). Low-velocity flow (7 cm/sec, a level similar to that experienced by the natural population in the Ohio River during summer and fall) was created by continuous operation of a small centrifugal water pump submersed in each tank. A larger pump ran continuously in the continuous-high velocity treatment, creating a 27 cm/sec flow (this fourfold increase above ambient velocities is similar to navigation-induced velocity increases that have been observed in potential habitats of riverine mussels, as discussed in detail in PART IV).

28. In the cyclic-high velocity treatment, the larger pump was activated for 5 min each hour with a programmable electronic timer. Water was maintained at 18° to 26° C and contained an ad libitum but nonfouling suspension of brewer's yeast for the duration of the 37-day experiment. Nutritionally adequate feeding of filter-feeding bivalves in a small closed system is not possible. The yeast suspension was provided for simplicity and because previous unpublished studies in our laboratory have shown that the yeast cells are ingested and used in partial support of maintenance metabolism.

Table 4

Means and Standard Deviations of Water Velocity Exposure, Tissue Condition Index, and Respiration Rate Measurements of Juvenile *F. ebena* in Three Velocity Exposure Treatments

Variable	Velocity Exposure Treatment		
	Continuous Low	Cyclic High	Continuous High
Water velocity, cm/sec			
Low	7.11 ± 1.02[a]	6.60 ± 1.02[a]	
High		26.42 ± 1.27[a]	27.18 ± 3.56[a]
Tissue Condition Index (TCI)			
(TDM/SDM)* × 100	1.72 ± 0.19[a]	1.69 ± 0.30[a]	1.43 ± 0.27[b]
Percent reduction**	19.73 ± 8.39[a]	22.39 ± 13.84[a]	34.48 ± 12.50[b]
Respiration rate			
$\mu mol\ O_2 \cdot mg^{-1} \cdot hr^{-1}$	1.45 ± 0.27[a]	1.46 ± 0.55[a]	1.75 ± 0.58[a]

Note: Superscript letters a and b indicate which means were not significantly different at the 0.05 level using Duncan's Multiple Range Test.

* TDM = tissue dry mass; SDM = shell dry mass.

** Percent reduction is relative to the TCI of juvenile *F. ebena* fixed in the field upon collection on 27 August.

29. On days 33, 35, and 37, a batch of eight mussels was removed from each of the three treatments to measure respiration and tissue condition. Respiration was measured by incubating each mussel in a 300-ml jar of water overnight in the dark at 22° C. After incubation, a 60-ml aliquot was siphoned from each jar, and dissolved oxygen determinations were made on each aliquot by Winkler titration. Three blanks were tested with each batch to determine bacterial oxygen uptake. Following determination of respiration, soft tissue was removed from the shell, and all tissues and shells were dried for 48 hr at 65° C and separately weighed. A tissue condition index (TCI) was obtained by dividing tissue dry mass by shell dry mass, both in milligrams, and multiplying the quotient by 100. A batch of juveniles fixed in 12-percent neutral formalin upon collection on 27 August was treated in an identical manner to estimate initial TCI.

Results and Discussion

30. The TCI of juvenile *F. ebena* in the continuous-low and cyclic-high velocity treatments was 20 and 22 percent less than the TCI of field-fixed juveniles. Continuous exposure to high-velocity water caused a 34-percent reduction in TCI. Comparison of the mean TCI by Duncan's Multiple Range Test indicated that weight loss was not significantly different ($p < 0.05$) between continuous-low and cyclic-high velocity treatments, but weight loss was significantly less in these two treatments than in the continuous-high velocity group (Table 4). Respiration rates, measured in still water, did not differ significantly among mussels from the three treatments.

31. Sustained changes in hydrologic conditions are known to affect pumping and filtration rates of marine lamellibranchs. These molluscs are sensitive to changes in flow (Kirby-Smith 1972, Walne 1972) and to small differences in pressure between the inhalent and exhalent siphons (Hildreth 1976). In addition, differences in the shape of unionids can be attributed to hydrologic conditions (Van der Schalie 1941, Clarke 1982 and references cited therein). With respect to turbulence, Brown, Clark, and Gleissner (1938) observed that the degree of stunted growth in unionids from the western basin of Lake Erie was positively correlated to the extent of exposure to waves.

32. The present experiment demonstrated that juvenile *F. ebena* are not affected by 5-min exposure to high-velocity flow once per hour, a result

directly relevant to evaluating the environmental effects of commercial navigation traffic. Commercial traffic rates in the upper Mississippi River and Ohio River do not often exceed one tow per hour (personal observations). Thus, turbulence caused by routine traffic is not likely to deleteriously affect mussels. Conversely, at sites where barges are fleeted, towboats sometimes work essentially continuously (personal observations). Potential impacts to mussels by abrupt water velocity changes in fleeting areas need to be evaluated on a site-specific basis.

33. Discharge of the lower Ohio River varies widely on a seasonal basis such that the range of water velocities experienced by mussels in the field is greater than the range between low and high flows used in the laboratory study. Parmalee (1956) reported that *F. ebena* inhabit sites with "swift current," although the population providing animals for the present experiment thrives in a slight current during normal summer and fall flows (Miller, Payne, and Siemsen 1986). The extent to which *F. ebena* is representative of growth and physiology of other unionids in large rivers has not been investigated. However, previous workers (Parmalee 1956, Fuller 1977, Buchanan 1980) indicate that *F. ebena* was, and in many cases still is (Miller, Payne, Siemsen 1986), a major component of gravel bar communities in large waterways.

PART IV: DISCUSSION AND SUMMARY

Relation of Laboratory Studies to Natural Populations

34. The most difficult aspect of any laboratory experiment on physiological stress is to evaluate the results in relation to naturally occurring populations. The laboratory studies reported herein were not intended to mimic exactly the turbulence and suspended solids perturbations caused by navigation traffic that could be experienced by naturally occurring mussel population. Rather, the studies were designed to determine the nature of sublethal physiological effects on freshwater mussels of intermittent pulses of marked changes in turbulence and suspended solids conditions. Even if the laboratory studies had been perfect mimics of navigation-related increases in turbulence and turbidity, other factors (such as food availability) would still differ from natural conditions. Man-induced impacts to natural habitats cannot be reproduced in the laboratory. Therefore, the results of the laboratory studies cannot be used directly to quantitatively predict the specific impacts on natural populations of mussels.

35. Most commercially valuable thick-shelled mussels are not in the main-channel habitats (as defined in Duncan and Thiel 1983); thus, the likelihood of their being exposed to intermittent turbulence and turbidity due to barge and tow passage is minimal. In the main channel, propeller wash is the most important source of turbulence. Theoretical models exist to predict levels of such turbulence at various depths and distances from the rotating propeller of a passing tow (e.g., Simons et al. 1981; Wuebben, Brown, and Zabilansky 1984). However, these models are unverified, and the physical forces generated by passing barges and tows in rivers are poorly understood (Simons et al. 1981; Wuebben, Brown, and Zabilansky 1984). Assuming general similarity in tow characteristics, the channel depth-to-tow draft ratio is the main determinant of how great near-bottom turbulence will be. Liou and Herbisch (1976, 1977) found that there was little movement of bed material when water depth-to-ship draft ratios were greater than two. Similarly, Fuller (1977) performed field studies of mussel distributions in the upper Mississippi River and concluded that mussels may be protected from mechanical damage by passing barges and tows when water depths exceed 6 m (greater than twice the draft of tows).

36. In the upper Mississippi River, main-channel border habitats support greater densities of mussels than the main channel (Duncan and Thiel 1983). In channel borders, the principal navigation-related sources of turbulence, and thus turbidity, are waves, drawdown, and surge caused by passing barges and tows (Simons et al. 1981). Several field studies have been conducted which involved attempts to quantify levels and durations of tow-induced turbulence or turbidity in channel borders (Johnson 1976, Bhowmik et al. 1981, Claflin et al. 1981, Eckblad 1981, Environmental Science and Engineering (ESE) 1981). Results of these studies were highly variable, and general conclusions that can be made are few. Reports of these studies have concluded that a crucial determinant of the level of turbulence created is the blocking ratio of a barge and tow (the ratio of the cross-sectional area of the barges and tow to the river channel). Turbulence and turbidity at any point on the bottom are also a function of the position of the actual sailing line, the water depth-to-ship draft ratio below this line, the type of bottom sediments below the sailing line and in adjacent areas, specific aspects of topography in the river's cross-sectional profile, and ambient patterns of turbulent flow and turbidity.

37. An instructive set of data on tow-induced changes in spatial and temporal patterns of flow near the bottom of the river in channel border habitats comes from field studies in Pool 9 of the upper Mississippi River and Pool 26 of the Illinois River (ESE 1981). These studies are directly relevant to the laboratory study discussed in PART III and are thus worthy of detailed description. In both rivers, barge and tow-induced changes in longshore and onshore water velocity vectors at nearshore and near-channel stations were recorded at 15-sec intervals using data loggers coupled to electromagnetic velocity probes positioned 1 ft (0.3 m) above the bottom at two stations.

38. In the Illinois River, ESE (1981) showed that barge and tow passage, on average, caused 8- to 18-cm/sec changes in the magnitude of longshore velocity vectors at both nearshore and near-channel monitoring stations. Barges and tows moving upstream generated a downstream increase in velocity, but traffic moving downstream forced velocity changes in the reverse direction. Because the ambient flow was only about 6 cm/sec, most downstream traffic caused a flow reversal at the monitoring stations. Longshore velocity changes were greater and in a consistent direction relative to onshore changes.

39. Results in the Mississippi River portion of the ESE study were more complex. Focusing on longshore velocity changes at the near-channel monitoring station, as in the Illinois River study, upbound tows caused ambient downstream currents at the near-channel station to increase; downbound tows had an opposite effect. On average, the maximum change in velocity was about 20 cm/sec, compared with an average ambient flow of about 25 cm/sec. However, nearshore changes in velocity were different than near-channel changes. Nearshore velocity patterns could not easily be interpreted with respect to barge and tow passage. Based on data in Appendix A of the ESE (1981) report, at least 8 of 23 barge and tow passage events could not be discerned from velocity readings at the nearshore station. Those measurements which showed a fairly clear relationship to barge and tow passage events showed that velocity changes at the nearshore station were opposite in direction and less in magnitude that those at the near-channel station. Nearshore velocities changed by an average of 10 cm/sec. Because ambient velocity at the nearshore station was generally near 0 cm/sec, upbound tows often caused brief upstream currents and downbound tows caused significant downstream currents. The duration of changes in nearshore or near-channel velocities averaged 1 to 2 min.

40. These field studies by ESE showed that barge and tow traffic could cause substantial intermittent changes in velocity conditions at shallow areas tens to hundreds of meters from the sailing line (on average, 180 and 75 m in the Illinois and Mississippi studies, respectively). The same studies also show that site-specific conditions determine to what extent, and even in what direction, velocity vectors may be changed. The level of velocity change in the second laboratory study reported herein falls within the range of changes observed in the field by ESE (1981).

41. Laboratory studies discussed in PART III showed that a 5-min increase in velocity of 18 cm/sec once per hour did not significantly reduce the tissue condition index of juvenile *F. ebena* relative to mussels continuously exposed to a velocity of 8 cm/sec. Barge and tow passage rates in the lower Ohio River, where *F. ebena* is the dominant unionid, generally do not exceed a rate of one passage per hour. Thus, the laboratory data suggest that *F. ebena* is not likely to be deleteriously affected by the velocity changes induced by routine traffic and likely to be experienced in channel border habitats where this species thrives (Miller et al., in preparation). However, both field variability in the effects of barge and tow traffic (as apparent in

the ESE data) and the general caution that should be taken when interpreting the results of laboratory experiments in the context of naturally occurring populations argue for field validation of these laboratory results.

42. The more complicated laboratory study of the combined effects of high levels of turbulence and turbidity (discussed in PART II) included a comparison of the degree of physiological stress experienced by mussels exposed to a 7-min burst of an unmeasured but quite high level of turbulence without turbidity once versus three times per hour. The more frequently exposed mussels showed evidence of some physiological stress (Table 1). Similarly, the laboratory study involving only velocity changes (PART III) showed that juvenile mussels exposed to a continuous flow of 25 cm/sec were stressed relative to mussels exposed to this level of velocity for 5 min each hour or continuously exposed to a velocity of 8 cm/sec (Table 4). Regardless of the potential for resuspension of bottom sediments, these effects of turbulence alone argue strongly for site-specific evaluations to ensure that barge fleeting areas are not permitted in areas close to mussel beds. In fleeting areas, tows sometimes work nearly continuously (personal observations), increasing the likelihood of sustained exposure of mussels to potentially deleterious levels of turbulence.

43. The laboratory results with respect to suspended solids exposure illustrate that high levels of turbidity can have an additive effect to turbulence. However, field studies of the effects of navigation on turbidity show that the levels of suspended solids (600 to 750 mg/ℓ) used in our laboratory attempts to elicit physiological stress responses will rarely be encountered by natural populations of mussels during periods of normal flow as a result of navigation traffic (Johnson 1976, Bhowmik et al. 1981, Claflin et al. 1981, Eckblad 1981).

44. The laboratory studies do suggest that disruption of normal feeding and therefore metabolic processes can be an important mechanism of physiological stress associated with exposure to high levels and frequencies of turbulence and suspended solids. Both laboratory studies suggested that shifts from food-based to body storage-based metabolism are associated with stressful conditions of turbulence and suspended solids exposure. Therefore, field studies directed at monitoring the effects of navigation traffic on natural beds of freshwater mussels should incorporate assessments of the growth condition (e.g., indices such as tissue-to-shell mass ratios) to provide early

warning of potential adverse impacts at the population or community level.

Suggested Field Studies

45. A critical link in evaluating potential adverse impacts of navigation traffic on naturally occurring mussels is to determine the turbulence and suspended solids exposure conditions of naturally occurring mussel beds. Main-channel border habitats and side-channel areas are the sites of the most diverse and dense assemblages of mussels in large navigable rivers such as the Mississippi and Ohio (Duncan and Thiel 1983). Field studies in such habitats should be conducted during low flow, when the physical effects of navigation can exceed natural levels of turbulence and turbidity. If significant increases in turbulence and turbidity are observed, potential impacts to mussels warrant further study to determine if the physical effects of traffic are adversely affecting the mussels. Site-specific studies offer the most direct means of evaluating navigation impacts to mussels.

46. If specific areas of a mussel bed are exposed to different levels of physical disturbance, it may be possible to compare the growth condition of individuals in different areas in an attempt to better define navigation impacts. Variables other than degree of exposure to the physical effects of navigation traffic could be involved. Therefore, in addition to comparisons between naturally occurring mussels in different areas, the use of manipulative experiments is often attractive. A useful manipulative experiment involves the caging of specimens of a single genetic stock in areas similar in all respects except exposure to navigation traffic. If navigation traffic patterns are expected to change over the years, long-term monitoring programs can be conducted to assess time trends in individuals' growth condition, population density and recruitment, and community composition in relation to changes in navigation traffic.

47. A difficult task in any monitoring effort is to acceptably define, a priori, what kinds and levels of biological effects constitute an unreasonably adverse change. Because so little is known about natural patterns of recruitment, growth, and mortality of freshwater mussels, criteria to define unreasonably adverse impacts will be difficult to agree upon. Unfortunately, without them, management decisions based on monitoring results can almost never be made.

48. With respect to changes in individual condition indices, it is important to recognize that unionids probably show large, natural, seasonal shifts in relation to reproductive periods and food availability. Many freshwater molluscs show wide seasonal shifts in growth condition. Mussels, because of their longevity and high fecundity, can be expected to show large seasonal shifts in tissue condition. Monitoring efforts, including measurements of growth condition of individuals, must be designed such that collections are made at the same time in the same season each year. For short-term, early-summer breeders (such as *F. ebena* in the lower Ohio River), fall is a season when the bioenergetic demands of spawning will have been largely overcome and mussels will have built up whatever energy reserves they need to successfully overwinter. In addition, fall is a season usually characterized by low water, so that sampling can be accomplished relatively easily.

Summary

49. Field studies have shown that some channel border habitats may be periodically exposed to changes in water velocity as a result of barge passage in the main channel. In the laboratory, the normal feeding mechanism of mussels is impaired by velocity changes of a magnitude within the range of changes that have been observed in some field studies. However, the brief episodes of impaired feeding that occur infrequently, such as those associated with routine navigation traffic, do not have a significant deleterious effect on the bioenergetic balance of individual mussels. Continuous disruption does have a significant effect, and mussels may be forced to depend on stored reserves when feeding impairment is sustained. Suspended solids exposure may be expected to have an additive effect to turbulence exposure.

50. Areas proposed for barge fleeting, where traffic can occur nearly continuously for sustained periods, must be evaluated carefully on a site-specific basis to ensure that valuable mussel populations are not in proximity. Site-specific studies should be performed to determine the frequency and magnitude of physical effects of navigation traffic that will be experienced by such populations. Physiological indices of stress (identical or similar to those used in the laboratory studies reported herein) can be used in field monitoring studies to provide an early warning of adverse biological impacts.

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